Ion Transport Investigation in Poplar Leaves by means of Nanofocussing refractive lenses and Micro-CT

[1]B. De Samber, [2]W. Schroeder, [3]C.G. Schroer, [4]G. Falkenberg and [1]L. Vincze

[1]X-ray Microspectroscopy and Imaging (XMI), Ghent University, Krijgslaan 281 – S12, B-9000 Ghent

[2]Forschungszentrum Juelich, 52425 Juelich

[3]Institute of Structural Physics, TU Dresden, D-01062 Dresden, Germany

[4]HASYLAB at DESY, Notkestrasse 85, D-22607 Hamburg

In a pilot study, poplar leaves were analysed at the P06 Hard X-ray micro/nanoprobe. The samples were shock-frozen in liquid propane followed by freeze-drying below the vitrification temperature of water. Fig. 1 shows the microscope image of the surface leave structure obtained by a Keyence light microscope installed at the beamline set-up. Several stomata openings are indicated; air containing carbon dioxide and oxygen enters the plant through these openings where it is used in photosynthesis and respiration, respectively. Ion transport plays an important role during stomatal opening.

X-ray fluorescence analysis was performed in order to gain insight in the elemental distribution of stomata in poplar leaves. Nanofocussing refractive lenses were used as focussing optics delivering an approximate beamsize of 80 x 80 nm^2 with a moderate flux of ~10^8 photons/s. Fig. 2 shows the elemental distribution of K (red), Ca (blue) and Zn (green) obtained on the poplar leaf (step size 1µm, 2s real time/point). A single opened stomata is marked by a white circle in the figure. Several Zn enriched spherical structures (green) can be observed, which are surrounded by a K-enriched layer (red). In between of these structures, a Ca-containing matrix is present homogeneously or as fibre-like structure (blue).

Fig. 3 (left) shows another scanned area on the poplar leaf with the same colour encoding. An interesting finding is the presence of a regular arrangement of Ca-crystals (encircled in white, presumably Ca-oxalate) associated to large vascular bundles. A single crystal was scanned in higher detail (200 nm step size, 4s real time/point) as shown in Fig. 3 (right). The coinciding presence of the vascular bundles with the Ca-crystals was confirmed by means of absorption micro-CT: Fig. 5 shows a single cross-section through such a vascular bundle showing similar trapezoid structures as found in the Ca-elemental distribution. The absorption micro-CT results were obtained via flat beam radiography (using a PCO4000 high resolution X-ray camera) under different rotation angles, followed by conventional backprojection algorithms. Rendering of the 3D data was performed by means of the OsiriX imaging software [1].

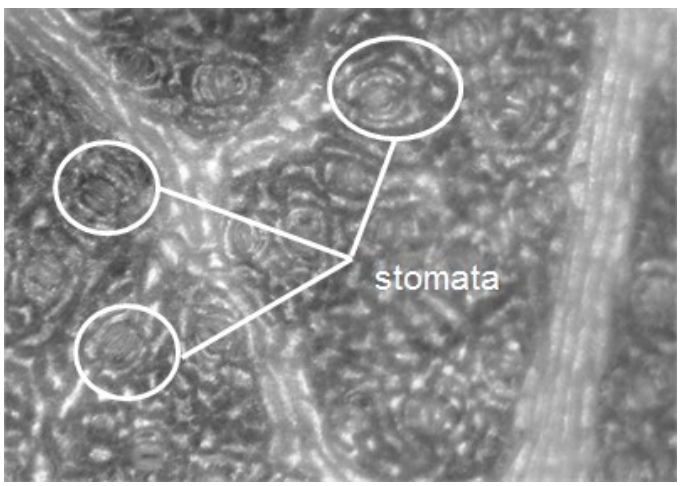

Fig1: microscope image of poplar leaf.

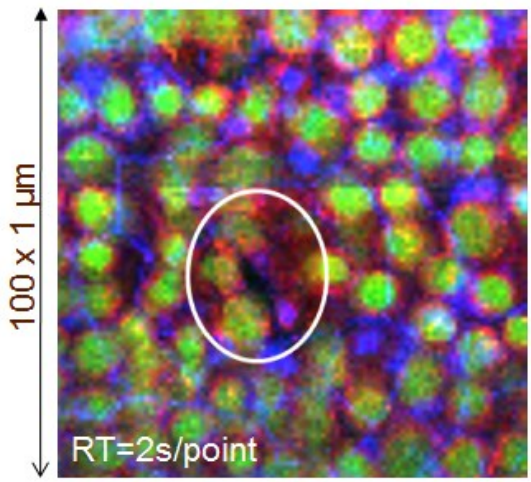

Fig. 2: Elemental distribution K, Ca and Zn.

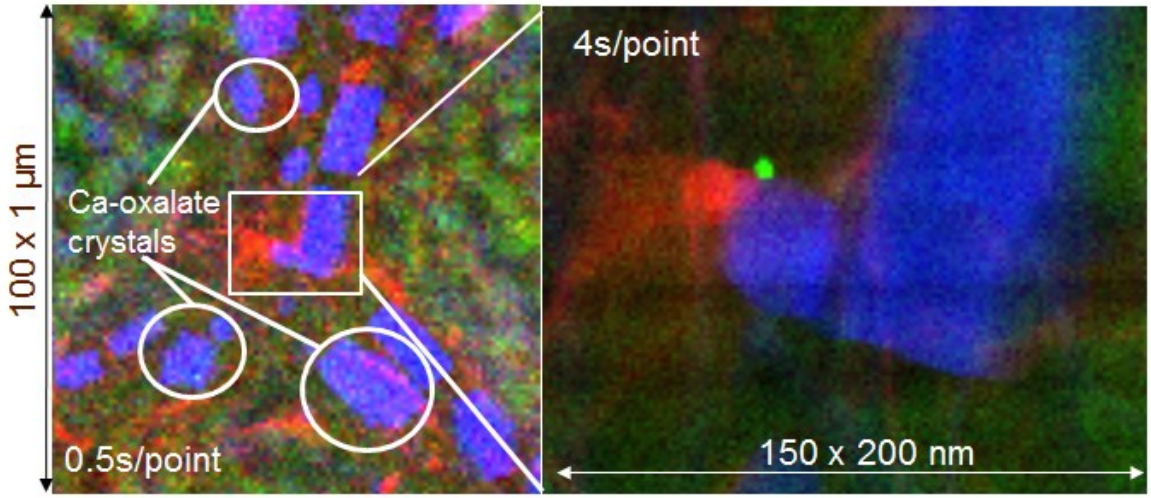

Fig. 3: elemental distributions of K, Ca and Zn in poplar leaf.

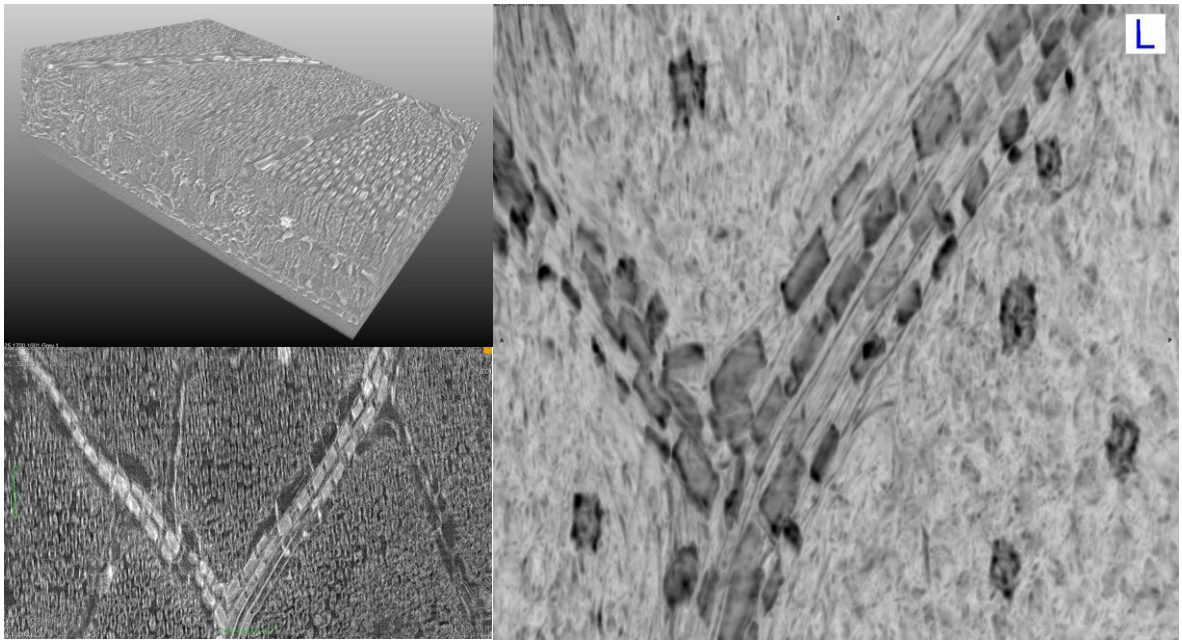

Fig. 4: absorption micro-CT of poplar leaf: 3D rendering (upper left), stack of 20 images (lower left) and single cross-section (right).

References

[1] http://www.osirix-viewer.com/,
OsiriX Imaging Software, Advanced Open-Source PACS Workstation DICOM Viewer.